DETAILED ACTION

1. The amendment and declaration filed on 4/1/2008 have been entered.

RCE Acknowledgment

2. A request for continued examination under 37 CFR 1.114, including the fee set forth in 37 CFR 1.17(e), was filed in this application after final rejection. Since this application is eligible for continued examination under 37 CFR 1.114, and the fee set forth in 37 CFR 1.17(e) has been timely paid, the finality of the previous Office action has been withdrawn pursuant to 37 CFR 1.114. Applicant's submission filed on 4/1/2008 has been entered.

3. Claims 57-65, 67-68, 71-76 are pending and are examined in the present office action. Claims 1-56, 66, 69-70 have been canceled.

4. Rejections and objections not set forth below are withdrawn.

Priority

5. The Office acknowledges Applicants' claim for domestic priority to application 09/738,398 filed 12/15/2000. Applicant is requested to amend the first paragraph to include the present status of said application.

Claim Objection

6. Claim 67 is objected to for being dependent on a canceled claim. For purposes of compact prosecution, claim 67 will still be examined as if it depends on claim 57 or 62.

Claim 65 is objected to under 37 CFR 1.75(c), as being of improper dependent form for failing to further limit the subject matter of a previous claim. Applicant is required to cancel the

claim(s), or amend the claim(s) to place the claim(s) in proper dependent form, or rewrite the claim(s) in independent form. The independent claim already states "regenerating a transformed plant from the stably transformed plant tissue or cells".

New Matter

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

7. Claims 57-65, 67-68, 71-76 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

The claims have been amended to recite "co-culturing on solid media monocot plant tissue or cells and an Agrobacterium…", "co-culturing on solid media plant tissue or cells and an Agrobacterium…" and "wherein if the sulfhydryl-containing agent is cysteine, cysteine is present at a concentration of 50 mg/L to 2000 mg/L". Applicants fail to point to support for the phrases in the instant specification. Upon a cursory search of the specification, support could not be found. Applicants are required to point to support for "co-culturing on solid media monocot plant tissue or cells and an Agrobacterium…", "co-culturing on solid media plant tissue or cells and an Agrobacterium…" and "wherein if the sulfhydryl-containing agent is cysteine, cysteine is

present at a concentration of 50 mg/L to 2000 mg/L" or to amend the claims to delete the NEW MATTER.

Enablement

8. Claims 57-61, 63-65, 67-68, 71-76 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention.

The claimed invention is not supported by an enabling disclosure taking into account the *Wands* factors. *In re Wands,* 858/F.2d 731, 8 USPQ2d 1400 (Fed. Cir. 1988). *In re Wands* lists a number of factors for determining whether or not undue experimentation would be required by one skilled in the art to make and/or use the invention. These factors are: the quantity of experimentation necessary, the amount of direction or guidance presented, the presence or absence of working examples of the invention, the nature of the invention, the state of the prior art, the relative skill of those in the art, the predictability or unpredictability of the art, and the breadth of the claim.

The claims are drawn to a method for the stable transformation of monocot plant tissue or cells comprising co-culturing on solid media monocot plant tissue or cells, with Agrobacterium containing a recombinant DNA, wherein the solid media comprises one or more sulfhydryl-containing agents, wherein said agent enhances stable transformation of the monocot plant tissue or cells relative to corresponding monocot plant tissue or cells without said agent, wherein if the sulfhydryl-containing agent is cysteine, cysteine is present at a concentration of 50 mg/L to 2000

mg/L, and regenerating a plant, or wherein transformation efficiency is at least 10% greater, or at least 0.5% greater, or enhanced by at least 5-fold, or wherein transformed tissue or cells are identified by a selectable or detectable marker, or wherein the plant tissue or cells are maize, wheat or rice tissue or cells, or wherein the plant tissue or cells are sugarcane.

Applicants have not reduced to practice their invention. Applicants claims are drawn to monocot plant tissue or cells but Applicants has not disclosed any method for transforming and regenerating any monocot plants from any tissue or cells. Applicants have not disclosed any media or transformation protocols that are specific to monocot plant tissues or cells. In experiments involving soybean, Applicants admit that Agrobacterium infection and the tissue culture process is genotype dependent (page 64, lines 6-8). In addition, Applicants disclose that for a method involving soybean, concentrations of cysteine between 800 mg/l and 1000 mg/l resulted in no growth of the hypocotyl (page 62, line 7). Applicants disclose that little callus growth was observed when using 1000 mg/l cysteine (page 66, lines 9-12). Applicants disclose that all levels of cysteine tested, except 50 mg/l, resulted in at least one explant with GUS positive shoot primordia (page 66, lines 23-24).

The state-of-the-art teaches that specific conditions and chemical components are required to achieve a successful transformation of a plant. Hansen et al (1999, Trends in Plant Science 4(6):226-231) teach that successful transformation of plants demands that certain criteria be met (page 227, under "Transformation systems"). Some of the requirements are that target tissues are competent for propagation or regeneration, an efficient DNA delivery method, and the ability to recover fertile transgenic plants at a reasonable frequency. Hansen et al also teach that there are variables that need to be tested to ensure success. These variables include the use of

feeder cells, alternative *Agrobacterium* strains, infiltration of the bacteria, and the duration and temperature of co-cultivation (page 228, right column, 3rd paragraph). Hansen also teaches that some crops appear to react or be hypersensitive to *Agrobacerium* and form necrotic barriers. To overcome this reaction, the addition of antioxidants is required (page 228, right column, last paragraph).

In the absence of guidance, undue trial and error experimentation would be required for one of ordinary skill in the art to test or evaluate the multitude of possible media formulations that may be effective for transforming and regenerating any monocot plants from monocot tissues or cells, and to select, if any, the transformed monocot plants that have been co-cultivated with any sulfhydryl-containing agents, or if the sulfhydryl-containing agent is cysteine, then the cysteine is present at a concentration of 50 mg/l to 2000 mg/l.

Therefore, given the breadth of the claims; the lack of guidance and examples; the unpredictability in the art; and the state-of-the-art as discussed above, undue experimentation would be required to practice the claimed invention, and therefore the invention is not enabled.

Claim Rejections - 35 USC § 102

(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

9. Claims 58-65, 67-68, 71-74 are rejected under 35 U.S.C. 102(b) as being anticipated by Perl et al (1996, Nature Biotechnology 14:624-628; listed in IDS).

The claims are drawn to a method for the stable transformation of plant tissue or cells comprising co-culturing on solid media plant tissue or cells and an Agrobacterium containing a recombinant DNA wherein the solid media comprises one or more sulfhydryl-containing agents

in an amount effective to enhance stable transformation of the tissue or cells compared to a control with the added agents, wherein if cysteine is present, it is at a concentration of 50 mg/L to 2000 mg/L, selecting transformed tissue and regenerating a whole plant, or wherein transformation is enhanced 10%, 0.5%, or 5-fold, or wherein the recombinant DNA comprises a selectable or detectable marker, or wherein the dithiothreitol (DTT) is present at between 0.75 to 2 mM.

Perl et al disclose a method for the stable transformation of grape comprising contacting embryogenic callus tissue and Agrobacterium containing recombinant DNA (page 627, left column, 3rd full paragraph) with the sulfhydryl-containing agent cysteine or DTT (page 625, Table 1), where these agent are present in a solid medium (page 625, left column, 1st full paragraph and Table 1), wherein the efficiency of stable transformation is enhanced by at least 5-fold, or at least 0.5% or 10% greater than in the absence of the agent (page 625, left column, 1st full paragraph; page 626, right column, last sentence of bottom full paragraph; page 627, left column, 1st full paragraph), where the transformed tissue is identified by hygromycin selection (page 626, Table 2), wherein stable transgenic grape plants are regenerated (Abstract, lines 8-9) and wherein the recombinant DNA contains a selectable detectable marker (page 626, Table 2; page 627, right column, "Selective culture"). Perl et al disclose the concentration of DTT is 0.2-5 mg/ml which is in the range of 0.75 to 2 mM as recited in claim 74 (the calculation was done using a DTT molecular weight of 154 g/mole).

Applicant's arguments filed 4/1/2008 have been fully considered but they are not persuasive.

Applicants contend that the only protocol disclosed in Perl et al to prepare transformed plants is the use of PVPP in solid co-cultivation medium followed by cultivation in a double-layer medium containing PVPP in the solid medium and DTT in the liquid medium (page 6 of Remarks, 3rd paragraph).

The Office contends that Perl et al disclose that DTT reduced browning to some extent and that the reduced browning of the embryogenic calli correlated with embryo regeneration (page 625, left column, 1st full paragraph). Perl et al state "Three percent of the embryogenic calli, cultured in medium supplemented with DTT, were able to produce de novo embryos" (*Ibid*).

In the Olhoft Declaration, Applicant summarizes some of the results from Perl et al and notes that the callus was transferred to a double layer medium with PVPP in the solid media and DTT in the upper (liquid) layer (page 2 of Declaration, paragraph 4). It is Applicant's opinion that the combination of PVPP and DTT in the double layer medium was likely chosen in experiments to produce stable transformation (page 2 of Declaration, paragraph 5). Applicants contend that while two sulfhydryl containing agents were tested in solid co-cultivation media in Perl et al (treatments 3 and 4 in Table 1), neither agent was chosen for the co-cultivation step in a protocol to prepare stable transformants (page 2 of Declaration, paragraph 6).

The Office contends that while Perl et al do disclose that the combination of PVPP and DTT was found to drastically inhibit necrogenesis of grape embryogenic calli following Agrobacterium co-cultivation (page 625, right column, top paragraph), Perl et al also state "By using tobacco leaf-disks as explants, we verified that the presence of PVPP and/or DTT during tobacco-Agrobacterium co-cultivation did not reduce the bacterial virulence" (page 625, right

column, 1st full paragraph). And Perl et al also disclose that DTT in a solid media did reduce necrogenesis of embryogenic calli (page 625, left column, 1st full paragraph and Table 1). The Office contends that even though the optimal procedure disclosed for transforming grape disclosed by Perl et al is not is not claimed in the instant application, Perl et al do disclose a procedure that is encompassed by the instantly claimed invention. Therefore, Perl et al anticipate Applicants' claimed invention.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

10. Claims 57-65, 67, 71-73 and 75-76 are rejected under 35 U.S.C. 103(a) as being unpatentable over Enriquez-Obregon et al (1997, Biotechnologia Aplicada 14(3):169-174) taken with Hansen et al (1999, Trends in Plant Science 4(6):226-231).

The claims are drawn to a method for the stable transformation of monocot plant tissue or cells or any plant tissue or cells comprising co-culturing on solid media monocot plant tissue or cells or co-culturing on solid media any plant tissue or cells, with Agrobacterium containing a recombinant DNA, wherein the solid media comprises one or more sulfhydryl-containing agents, wherein said agent enhances stable transformation of the monocot plant tissue or cells relative to corresponding monocot plant tissue or cells or any plant tissue or cells without said agent,

wherein if the sulfhydryl-containing agent is cysteine, cysteine is present at a concentration of 50 mg/L to 2000 mg/L, and regenerating a plant, or wherein transformation efficiency is at least 10% greater, or at least 0.5% greater, or enhanced by at least 5-fold, or wherein transformed tissue or cells are identified by a selectable or detectable marker, or wherein the plant tissue or cells are maize, wheat or rice tissue or cells, or wherein the plant tissue or cells are sugarcane.

Enriquez-Obregon et al teach a method for the stable transformation of the monocot sugarcane comprising contacting meristematic stem tissue and Agrobacterium containing recombinant DNA (page 170, Figure 1) with the sulfhydryl-containing agent cysteine (page 170, Table 1), where the cysteine was present in a solid medium at a concentration of 40 mg/L (page 172, left column, 1st full sentence and page 170, Table 1). Enriquez-Obregon et al disclose explants in contact with cysteine containing media had a greater transformation efficiency than those explants not grown on media containing cysteine (page 171, Table 3). The results of Enriquez-Obregon et al demonstrate a 10%, 0.5% and greater than 5-fold increase. Enriquez-Obregon et al disclose the transformed explants were identified by a selectable and detectable marker, i.e., GUS (for example see page 171, Table 3), or wherein a recombinant DNA contains selectable detectable markers (page 170 Figure 1). Enriquez-Obregon et al teach the regeneration of stable Agrobacterium transformed transgenic sugarcane plants. (page 174, right column, bottom paragraph).

Sugarcane is a monocot plant. The Agrobacterium used contained maize ubiquitin promoter-BarR coding sequence DNA and a CaMV 35S promoter operably linked to a uidA (GUS) coding sequence. This DNA functions as a selectable marker for Bar-resistance, as well as a detectable marker, since the uidA (GUS) coding sequence expression allows visualization by causing the development of blue color under the proper conditions. The sulfhydryl-containing agent cysteine was present in solid medium with the Agrobacterium/ plant material (page 172, left column, 1st full sentence).

Enriquez-Obregon et al do not teach a co-cultivation solid media comprising a cysteine concentration of between 50 mg/L to 2000 mg/L or transformation of maize, wheat or rice tissue or cells.

Enriquez-Obregon et al do teach a liquid medium comprising 90 mg/L cysteine in which Agrobacterium and explants were placed (page 171, Table 2).

Given the recognition of those of ordinary skill in the art the value of transforming a sugarcane plant to improve the plant's agricultural yields and industrial production as taught by Enriquez-Obregon et al (page 169, left column, 1st paragraph), one skilled in the art would be motivated to use the method of Enriquez-Obregon et al for transforming sugarcane and to optimize process parameters by varying the cysteine concentration to be between 50 mg/L and 2000 mg/L and to include the cysteine in a solid media, absent evidence to the contrary. One skilled in the art would also be motivated to use the method of Enriquez-Obregon et al for the transformation of other monocot plants, such as maize, wheat or rice, given the teachings of Hansen et al which state "To accommodate a genotype or species that has not been manipulated in culture previously, one must either adapt an established protocol or create a new one…".

Therefore, one skilled in the art would be motivated to use the method of Enriquez-Obregon et al for transformation of other monocot species such as maize, wheat or rice.

Thus the claimed invention would have been *prima facie* obvious as a whole to one of ordinary skill in the art at the time it was made, especially in the absence of evidence to the contrary.

Applicant's arguments filed 4/1/2008 have been fully considered but they are not persuasive.

In the Amendment filed on January 7, 2008, Applicant traversed the assertion that "one of ordinary skill in the art recognizes that a transformation procedure that works for one member of a group will also work for other members of the group" as a form of Official Notice and requested a reference to support the assertion or an affidavit of personal knowledge by the Examiner (page 7 of Remarks, 1st full paragraph).

The Office contends that "Official Notice" was not taken because it was not stated explicitly. The present 103 rejection includes a reference by Hansen et al that supports the Office's assertion.

In the Olhoft Declaration, Applicant contends there is a balance between plant cell viability and agent toxicity (page 3 of Declaration, paragraph 10). Applicant notes that for experiments to test the effect of the combination of agents on Agrobacterium-mediated gene transfer, Enriquez-Obregon et al selected the lower concentration of the two tested concentrations for use in the pre-coculture liquid medium and co-culture medium (page 3 of Declaration, paragraph 10).

The Office contends that while Enriquez-Obregon et al used the lower concentration of the two tested concentrations of each of the three agents in their medium, the references taken together still make obvious Applicants' claimed invention. See In re Kuhle, 188 USPQ 7, (CCPA 1975), which teaches that a feature which solves no stated problem and which presents no unexpected results would have been an obvious matter of choice.

11. No claims are allowed.

12. Any inquiry concerning this communication or earlier communications from the examiner should be directed to Stuart F. Baum whose telephone number is 571-272-0792. The examiner can normally be reached on M-F 8:30-5:00.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Anne Marie Grunberg can be reached at 571-272-0975. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Any inquiry of a general nature or relating to the status of this application or proceeding should be directed to the receptionist whose telephone number is 571-272-1600.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

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